



## **Diversity of total and functional microbiome of anammox reactors fed with complex and synthetic nitrogen-rich wastewaters**

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*Total number of authors:*

11

*Publication date:*

2013

*Document Version*

Publisher's PDF, also known as Version of record

[Link back to DTU Orbit](#)

*Citation (APA):*

Gülay, A., Pellicer i Nàcher, C., Mutlu, A. G., Jensen, M. M., Vlaeminck, S., Lackner, S., Terada, A., Sørensen, S. J., Hansen, L., Al-Soud, W., & Smets, B. F. (2013). *Diversity of total and functional microbiome of anammox reactors fed with complex and synthetic nitrogen-rich wastewaters*. Abstract from ICON3: 3rd international conference on Nitrification, Tokoy, Japan.

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**Category:** Advanced Molecular ecology

**Title:** Diversity of total and functional microbiome of anammox reactors fed with complex and synthetic nitrogen-rich wastewaters

**Keyword:** pyrosequencing, anammox, substrate complexity

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**Abstract:**

There are few comparative studies of microbial structure, composition and phylogenetic diversity of the anammox reactors as a function of substrate complexity exist, representing a large gap in the scientific literature.

In this study, we applied 16S rRNA gene (rDNA) tag-based 454 pyrosequencing as a deep sequencing approach to 59 biomass samples from 24 different anammox bioreactors together with proper biological replication in order to compare their total and functional (wrt anaerobic ammonium oxidation) microbial diversity. Among 24 sampled bioreactors, 10 of them were full scale implementations treating complex nitrogen-rich wastewaters and 14 were lab-scale implementations treating synthetic wastewaters. We found that nitrification/anammox bioreactors treating complex nitrogen-rich wastewaters were more diverse in terms of total microbial diversity but less diverse at anammox functional diversity than the bioreactors treating synthetic wastewaters inferred from observed OTUs, Chao1, Shannon index and Phylogenetic distance calculations. Differences in total microbial diversity agreed with the ecological theory concerning the positive correlation between substrate complexity and biodiversity (Parrott 2010), but, not (Harris et al. 2012) in the anammox functional guild diversity (functional diversity term was used based on phylogenetic groups known to harbor the anammox metabolic pathway). Classifying the microbial structure of bioreactors according to substrate complexity using weighted UniFrac algorithm explained 29% of the variance where the bioreactor samples of complex nitrogen-rich wastewater feeding was clearly separated from the bioreactor samples of synthetic feeding.

Here we examined and compared for the first time microbial diversity of nitrification-anammox reactors that are designed and built individually for treating complex nitrogen-rich and synthetic wastewaters across the world using 16 rRNA gene pyrosequencing. With the aid of replicated genetic snapshots, we revealed the relationship between the microbial diversity of nitrification-anammox reactors operated by different substrate complexity in terms of microbial composition, structure, richness and phylogenetic diversity from two points of view: total and functional diversity.